

(Fig.3 pg 1 of 3)

Figure 3: Kits for discovery of, or application of diagnostic gene sets

A. Contents of kit for discovery of diagnostic gene sets using microarrays

1. Sterile, endotoxin and RNase free blood collection tubes
2. Alcohol swabs, tourniquet, blood collection set
- 3.-PBS (phosphate buffer saline; needed when method of example 8 is used to derived mononuclear RNA)
4. Cell lysis buffer
5. RNA isolation kit
6. Substrates for labeling of RNA (may vary for various expression profiling techniques)
 - For fluorescence microarray expression profiling:
 - Reverse transcriptase and 10x RT buffer
 - T7(dT)24 primer (primer with T7 promoter at 5' end)
 - DTT
 - Deoxynucleotides 100mM each
 - RNase inhibitor
 - 2nd strand cDNA buffer
 - DNA polymerase
 - Rnase H
 - T7 RNA polymerase
 - Ribonucleotides
 - In Vitro transcription buffer
 - Cy3 and Cy5 labeled ribonucleotides
7. Microarrays containing candidate gene libraries
8. Cover slips for slides
9. Hybridization chambers
10. Software package for identification of diagnostic gene set from data
 - Contains statistical methods.
 - Allows alteration in desired sensitivity and specificity of gene set.
 - Software facilitates access to and data analysis by centrally located database server.
11. Password and account number to access central database server.
12. Kit User Manual

B. Contents of kit for application of diagnostic gene sets using microarrays

1. Sterile, endotoxin and RNase free blood collection tubes
2. Alcohol swabs, tourniquet, blood collection set
- 3.-PBS (phosphate buffer saline; needed when method of example 7 is used to derived mononuclear RNA)

(Fig.3 pg 2 of 3)

4. Cell lysis buffer
5. RNA isolation kit
6. Substrates for labeling of RNA (may vary for various expression profiling techniques)
 - For fluorescence microarray expression profiling:
 - Reverse transcriptase and 10x RT buffer
 - T7(dT)24 primer (primer with T7 promoter at 5' end)
 - DTT
 - Deoxynucleotides 100mM each
 - RNAse inhibitor
 - 2nd strand cDNA buffer
 - DNA polymerase
 - Rnase H
 - T7 RNA polymerase
 - Ribonucleotides
 - In Vitro transcription buffer
 - Cy3 and Cy5 labeled ribonucleotides
7. Microarrays containing candidate gene libraries
8. Cover slips for slides
9. Hybridization chambers
10. Software package for identification of diagnostic gene set from data
 - Contains statistical methods.
 - Allows alteration in desired sensitivity and specificity of gene set.
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C. Contents of kit for application of diagnostic gene sets using Real-time RT-PCR

1. Sterile, endotoxin and RNAse free blood collection tubes
2. Alcohol swabs, tourniquet, blood collection set
3. PBS (phosphate buffer saline; needed when method of example 7 is used to derived mononuclear RNA)
4. Cell lysis buffer
5. RNA isolation kit
6. Substrates for real time RT-PCR (may vary for various real-time PCR techniques:
 - poly dT primers, random hexamer primers
 - Reverse Transcriptase and RT buffer
 - DTT
 - Deoxynucleotides 100 mM
 - RNAse H
 - primer pairs for diagnostic and control gene set
 - 10x PCR reaction buffer

(Fig.3 pg 3 of 3)

- Taq DNA polymerase
- Fluorescent probes for diagnostic and control gene set
(alternatively, fluorescent dye that binds to only double stranded DNA)
- reaction tubes with or without barcode for sample tracking
- 96-well plates with barcode for sample identification, one barcode for entire set,
or individual barcode per reaction tube in plate
- 7. Software package for identification of diagnostic gene set from data
 - Contains statistical methods.
 - Allows alteration in desired sensitivity and specificity of gene set.
 - Software facilitates access to and data analysis by centrally located database
server
- 8. Password and account number to access central database server.
- 9. Kit User Manual